APPENDIX II

AFFIDAVIT BY DR. SUSAN DAGHER

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re United States Patent Application of:		**Docket No.:**	**4176-101**
Applicant:	**CHILKOTI, ASHUTOSH**	**Examiner:**	**Walicka, M.A.**
Application No.:	**09/812,382**	**Art Group:**	**1652**
Date Filed:	**March 20, 2001**	**Confirm. No.:**	**1286**
Title:	**FUSION PEPTIDES ISOLATABLE BY PHASE TRANSITION**		

23448

EXPRESS MAIL CERTIFICATE

I hereby certify that I am mailing the attached documents to the Commissioner for Patents on March 1, 2004 in an envelope addressed to the Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450, and Express Mailed under the provisions of 37 CFR 1.10.

L. Stephen Lockett

March 1, 2004
Date

EV160595225US
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AFFIDAVIT UNDER 37 C.F.R. §1.132 OF DR. SUSAN DAGHER IN U.S. PATENT APPLICATION NO. 09/812,383

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

I, **DR. SUSAN DAGHER**, being duly sworn, depose and say:

(1) THAT I have a PhD in molecular biology and am currently working as the primary research investigator for Phase BioScience, Inc (Chapel Hill, North Carolina), the licensee of U.S. Patent Application No. 09/812,383, filed on March 20, 2001 in the U.S. Patent and Trademark Office in my name for "**FUSION PEPTIDES ISOLATABLE BY PHASE TRANSITION**" (hereinafter "the Application"), which claims priority to U.S. Provisional Patent Application No. 60/190,659 filed March 20, 2000.

(2) THAT the Application discloses and claims fusion proteins that each comprise one or more target proteins of interest fused with one or more elastin-like polypeptides (ELPs) exhibiting inverse phase transition behavior, while such fusion proteins retain the inverse phase transition behavior of the ELPs and therefore can be isolated from other soluble proteins by inverse transition cycling (ITC) process (hereinafter "the Invention").

(3) THAT in support of the Application, experiments have been conducted to show the use of various target proteins in forming ELP-containing fusion proteins and the inverse phase transition behavior exhibited by such fusion proteins. Specifically, thirty-six (36) ELP-containing fusion proteins were formed in *E. coli* by using known recombinant expression techniques consistent with the teachings and disclosures in Sections 5.4 and 6 of the Application.

(4) THAT the thirty-six ELP-containing fusion proteins comprised the combinations of:

- Insulin A peptide and ELP4-60 polypeptide with an enterokinase protease cleavage site therebetween;
- Insulin A peptide and ELP1-90 polypeptide with an enterokinase protease cleavage site therebetween;

- Insulin A peptide and ELP4-120 polypeptide with an enterokinase protease cleavage site therebetween;
- Insulin A peptide and ELP1-180 polypeptide with an enterokinase protease cleavage site therebetween;
- T20 peptide and ELP4-60 polypeptide with an enterokinase protease cleavage site therebetween;
- T20 peptide and ELP1-90 polypeptide with an enterokinase protease cleavage site therebetween;
- T20 peptide and ELP4-120 polypeptide with an enterokinase protease cleavage site therebetween;
- T20 peptide and ELP4-60 polypeptide with a thrombin protease cleavage site therebetween;
- T20 peptide and ELP1-90 polypeptide with a thrombin protease cleavage site therebetween;
- T20 peptide and ELP4-120 polypeptide with a thrombin protease cleavage site therebetween;
- T20 peptide and ELP4-60 polypeptide with a tobacco etch virus (TEV) protease cleavage site (cleavage between QS residues) therebetween;
- T20 peptide and ELP1-90 polypeptide with a TEV protease cleavage site (cleavage between QS residues) therebetween;
- T20 peptide and ELP4-120 polypeptide with a TEV protease cleavage site (cleavage between QS residues) therebetween;
- T20 peptide and ELP4-60 polypeptide with a TEV protease cleavage site (cleavage between QY residues) therebetween;
- T20 peptide and ELP1-90 polypeptide with a TEV protease cleavage site (cleavage between QY residues) therebetween;
- T20 peptide and ELP4-120 polypeptide with a TEV protease cleavage site (cleavage between QY residues) therebetween;

- Interferon alpha 2B protein and ELP1-90 polypeptide with a thrombin protease cleavage site therebetween;
- Tobacco etch virus protease and ELP4-60 polypeptide with a thrombin protease cleavage site therebetween;
- Tobacco etch virus protease and ELP1-90 polypeptide with a thrombin protease cleavage site therebetween;
- Tobacco etch virus protease and ELP4-120 polypeptide with a thrombin protease cleavage site therebetween;
- Tobacco etch virus protease and ELP1-180 polypeptide with a thrombin protease cleavage site therebetween;
- Small heterodimer partner orphan receptor and ELP1-90 polypeptide with a thrombin protease cleavage site therebetween;
- Androgen receptor ligand binding domain and ELP1-90 polypeptide with a thrombin protease cleavage site therebetween;
- Androgen receptor ligand binding domain and ELP1-180 polypeptide with a thrombin protease cleavage site therebetween;
- Glucocorticoid receptor ligand binding domain and ELP1-90 polypeptide with a thrombin protease cleavage site therebetween;
- Estrogen receptor ligand binding domain and ELP1-60 polypeptide with a thrombin protease cleavage site therebetween;
- Estrogen receptor ligand binding domain and ELP1-90 polypeptide with a thrombin protease cleavage site therebetween;
- Estrogen receptor ligand binding domain and ELP1-180 polypeptide with a thrombin protease cleavage site therebetween;

- Estrogen receptor ligand binding domain and ELP1-90 polypeptide with a TEV protease cleavage site (cleavage between QG residues) therebetween;
- G protein alpha Q and ELP1-90 polypeptide with a thrombin protease cleavage site therebetween;
- G protein alpha Q and ELP1-180 polypeptide with a thrombin protease cleavage site therebetween;
- 1-Deoxy-D-Xylulose 5-Phosphate reductoisomerase peptide and ELP1-60 polypeptide with a thrombin protease cleavage site therebetween;
- 1-Deoxy-D-Xylulose 5-Phosphate reductoisomerase peptide and ELP1-90 polypeptide with a thrombin protease cleavage site therebetween;
- 1-Deoxy-D-Xylulose 5-Phosphate reductoisomerase peptide and ELP1-180 polypeptide with a thrombin protease cleavage site therebetween;
- 1-Deoxy-D-Xylulose 5-Phosphate reductoisomerase peptide and ELP1-90 polypeptide with a TEV protease cleavage site (cleavage between QG residues) therebetween; and
- G protein alpha S and ELP1-90 polypeptide with a thrombin protease cleavage site therebetween.

(See **Appendix A** and Section D of **Appendix B** enclosed herewith)

(5) THAT a total of eleven (11) different target proteins were used for forming the ELP-containing fusion proteins as listed in paragraph (4) above, which included:

(a) Insulin A peptide comprising 21 amino acid residues with molecular weight of about 2,400 Daltons;

(b) T20 peptide comprising 36 amino acid residues with molecular weight of about 4,400 Daltons;

(c) Interferon alpha 2B peptide comprising 188 amino acid residues with molecular weight of about 21,500 Daltons;

(d) Tobacco etch virus protease comprising 242 amino acid residues with molecular weight of about 27,500 Daltons;

(e) Small Heterodimer partner orphan receptor comprising 257 amino acid residues with molecular weight of about 28,000 Daltons;

(f) Androgen receptor ligand binding domain comprising 258 amino acid residues with molecular weight of about 30,100 Daltons;

(g) Glucocorticoid receptor ligand binding domain comprising 279 amino acid residues with molecular weight of about 32,100 Daltons;

(h) Estrogen receptor ligand binding domain comprising 296 amino acid residues with molecular weight of about 33,200 Daltons;

(i) G protein alpha Q comprising 359 amino acid residues with molecular weight of about 42,100 Daltons;

(j) 1-Deoxy-D-Xylulose 5-Phosphate reductoisomerase peptide comprising 400 amino acid residues with molecular weight of about 43,5 Daltons; and

(k) G protein alpha S comprising 380 amino acid residues with molecular weight of about 44,200 Daltons.

(See the target protein sequences in Section A of **Appendix B** enclosed herewith.)

(6) THAT the eleven target proteins as listed in paragraph (5) hereinabove are different in their respective:

- primary structures;
- secondary structures;
- tertiary structures;

- molecular weights;
- electric charge distributions;
- viscosity; and
- biological functions.

(7) THAT five (5) different elastin-like polypeptides (ELPs) were used for forming the fusion proteins as listed in paragraph (4) hereinabove. The sequences of such ELPs are disclosed in Section B of **Appendix B** enclosed herewith.

(8) THAT eleven (11) different spacer peptides containing various protease cleavage sites were used for joining the target proteins with the ELPs in forming the fusion proteins as listed in paragraph (4) hereinabove. The spacer peptide sequences are disclosed in Section C of **Appendix B** enclosed herewith.

(9) THAT all of the thirty-six ELP-containing fusion proteins as listed in paragraph (4) hereinabove retained the inverse phase transition behavior of the ELPs.

(10) THAT the ELP-containing fusion proteins as listed in paragraph (4) hereinabove were specifically isolated and purified by using inverse transition cycling (ITC) techniques, according to the following experimental procedure:

(A) Isolation and Purification of Fusion Proteins Containing Insulin A Peptide (InsA)

A single colony of *E. coli* strain BLR (DE3) (Novagen) containing the respective ELP-InsA fusion protein was inoculated into 5 ml CircleGrow (Q-BIOgene, San Diego, CA) supplemented with 100 μg/ml ampicillin (Sigma) and grown at 37°C with shaking at 250 rpm for 5 hours. The

5 ml culture was then inoculated into a 500 ml culture and allowed to grow at 25°C for 16 hours before inducing with 1 mM IPTG for 4 hours at 25°C. The culture was harvested and suspended in 40 ml 20 mM Tris-HCL pH 7.4, 50 mM NaCl, 1 mM DTT and 1 Complete EDTA free Protease inhibitor pellet (Roche, Indianapolis, IN). Cells were lysed by ultrasonic disruption on ice for 3 minutes, which consisted of 10 seconds bursts at 35% power separated by 30 second cooling down intervals. Cell debris was removed by centrifugation at 20,000g, 4°C for 30 minutes.

Inverse phase transition was induced by adding NaCl to the cell lysate at room temperature to achieve a final concentration of 1.0 M therein, followed by centrifugation at 20,000g for 15 minutes at room temperature. The resulting pellet contained the respective ELP-InsA fusion protein and non-specifically NaCl precipitated proteins.

The pellet was re-suspended in 40 ml ice-cold ml 20 mM Tris-HCL pH 7.4, 50 mM NaCl, 1 mM DTT and re-centrifuged at 20,000 g, 4°C for 15 minutes to remove the non-specifically NaCl precipitated proteins. The inverse transition cycle was repeated two additional times to increase the purity of the respective ELP-InsA fusion protein and reduce the final volume to 0.5 ml.

(B) Isolation and Purification of Fusion Proteins Containing T20 Peptide (T20)

A single colony of *E. coli* strain BLR (DE3) (Novagen) containing the respective ELP-T20 fusion protein was inoculated into 500 ml CircleGrow (Q-BIOgene, San Diego, CA) supplemented with 100 μg/ml ampicillin (Sigma) and grown at 37°C with shaking at 250 rpm for 24 hours. The culture was harvested and suspended in 40 ml 50 mM Tris pH 8.0, 0.5 mM EDTA and 1 Complete Protease inhibitor pellet (Roche, Indianapolis, IN). Cells were lysed by ultrasonic disruption on ice for 3 minutes, which consisted of 10 seconds bursts at 35% power

separated by 30 second cooling down intervals. Cell debris was removed by centrifugation at 20,000g, 4°C for 30 minutes.

Inverse phase transition was induced by adding NaCl to the cell lysate at room temperature to achieve a final concentration of 1.0 M therein, followed by centrifugation at 20,000g for 15 minutes at room temperature. The resulting pellet contained the respective ELP-T20 fusion protein and non-specifically NaCl precipitated proteins.

The pellet was re-suspended in 40 ml ice-cold ml 50 mM Tris pH 8.0, 0.5 mM EDTA and re-centrifuged at 20,000 g, 4°C for 15 minutes to remove the non-specifically NaCl precipitated proteins. The inverse transition cycle was repeated two additional times to increase the purity of the respective ELP-T20 fusion protein and reduce the final volume to 5 ml.

(C) Isolation and Purification of Fusion Protein Containing Interferon Alpha 2B Peptide (IFNA2)

A single colony of *E. coli* strain BL21(DE3) TrxB$^-$ (Novagen) containing the ELP-IFNA2 fusion protein and Codon Plus-RIL plasmid (Stratagene) was inoculated into 500 ml CircleGrow (Q-BIOgene, San Diego, CA) supplemented with 100 μg/ml ampicillin (Sigma), 25 ug/ml Chloramphenicol (Sigma) and incubated at 27°C with shaking at 250 rpm for 48 hours. The culture was harvested and suspended in 50 mM Tris-HCL pH 7.4, 50 mM NaCl and 1 Complete EDTA free Protease inhibitor pellet (Roche, Indianapolis, IN). Cells were lysed by ultrasonic disruption on ice for 3 minutes, which consists of 10 seconds bursts at 35% power separated by 30 second cooling down intervals. Cell debris was removed by centrifugation at 20,000g, 4°C for 30 minutes.

Inverse phase transition was induced by adding NaCl to the cell lysate at room temperature to achieve a final concentration of 1.5 M, followed by centrifugation at 20,000g for 15 minutes at room temperature. The resulting pellet contained the ELP-IFNA2 fusion protein and non-specifically NaCl precipitated proteins.

The pellet was re-suspended in 40 ml ice-cold 50 mM Tris-HCL pH 7.4 and 50 mM NaCl and re-centrifuged at 20,000 g, 4°C for 15 minutes to remove the non-specifically NaCl precipitated proteins. The inverse transition cycle was repeated two additional times to increase the purity of the ELP-IFNA2 fusion protein and reduce the final volume to 5 ml.

(D) Isolation and Purification of Fusion Proteins Containing Tobacco Etch Virus Protease (TEV)

A single colony of *E. coli* strain BL21 star or BRL(DE3) containing pET15b-SD5-ELP-TEV constructs and Codon Plus-RIL plasmid (Stratagene) was inoculated into 500 ml CircleGrow (Q-BIOgene, San Diego, CA) supplemented with 100 µg/ml ampicillin (Sigma), 25 ug/ml Chloramphenicol (Sigma) and incubated at 27°C with shaking at 250 rpm for 48 hours. The culture was harvested and suspended in 50 mM Tris-HCL pH 8.0, 1 mM EDTA, 5 mM DTT, 10% glycerol and 1mM PMSF. Cells were lysed by ultrasonic disruption on ice for 3 minutes, consisting of 10 seconds bursts at 35% power separated by 30 second cooling down intervals. Cell debris was removed by centrifugation at 20,000g, 4°C for 30 minutes.

Inverse phase transition was induced by adding NaCl to the cell lysate at room temperature to achieve a final concentration of 1.5 M, followed by centrifugation at 20,000g for 15 minutes at room temperature. The resulting pellet contained the respective ELP-TEV fusion protein and non-specifically NaCl precipitated proteins.

The pellet was re-suspended in 40 ml ice-cold 50 mM Tris-HCL pH 8.0, 1 mM EDTA, 5 mM DTT, 10% glycerol and re-centrifuged at 20,000 g, 4°C for 15 minutes to remove the non-specifically NaCl precipitated proteins. The inverse transition cycle was repeated two additional times to increase the purity of the respective ELP-TEV fusion protein and reduce the final volume to 1 ml.

(D) Isolation and Purification of Fusion Protein Containing Small Heterodimer Partner Orphan Receptor (SHP)

A single colony of *E. coli* strain BL21 Star (DE3) containing the ELP-SHP fusion protein was inoculated into 500 ml CircleGrow (Q-BIOgene, San Diego, CA) supplemented with 100 μg/ml ampicillin (Sigma) and 10% sucrose and grown at 27°C with shaking at 250 rpm for 48 hours. The culture was harvested and suspended in 50 mM Tris-HCL pH 8.0, 150 mM KCL, 1 mM DTT 1 mM EDTA and 1 Complete EDTA free Protease inhibitor pellet (Roche, Indianapolis, IN). Cells were lysed by ultrasonic disruption on ice for 3 minutes, which consistes of 10 seconds bursts at 35% power separated by 30 second cooling down intervals. DNA and RNA in the soluble lysate were further degraded by adding 2 μl Benzonase (Novagen) and incubating at 4°C for 30 minutes. Cell debris was removed by centrifugation at 20,000g, 4°C for 30 minutes.

Inverse phase transition was induced by adding NaCl to the cell lysate at room temperature to achieve a final concentration of 1.5 M, followed by centrifugation at 20,000g for 15 minutes at room temperature. The resulting pellet contained the ELP-SHP fusion protein and non-specifically NaCl precipitated proteins.

The pellet was re-suspended in 40 ml ice-cold 50 mM Tris-HCL pH 8.0, 150 mM KCL, 1 mM DTT 1 mM EDTA, and 1% N-Octylglucoside and re-centrifuged at 20,000 g, 4°C for 15 minutes

to remove non-specific insoluble proteins. The temperature transition cycle was repeated two additional times to increase the purity of the ELP-SHP fusion protein and reduce the final volume to 2 ml.

(F) Isolation and Purification of Fusion Proteins Containing Androgen Receptor Ligand Binding Domain (AR-LBD)

A single colony of *E. coli* strain BL21 Star (DE3) containing the respective ELP-AR-LBD fusion protein was inoculated into 500 ml CircleGrow (Q-BIOgene, San Diego, CA) supplemented with 100 μg/ml ampicillin (Sigma) and 10 μM DHT and grown at 27°C with shaking at 250 rpm for 48 hours. The culture was harvested and suspended in 40 ml 50mM Hepes pH 7.5, 150 mM NaCl, 0.1% N-Octylglycoside, 10% glycerol, 1 mM DTT, 1 μM DHT and 1 Complete EDTA free Protease inhibitor pellet (Roche, Indianapolis, IN). Cells were lysed by ultrasonic disruption on ice for 3 minutes, which consisted of 10 seconds bursts at 35% power separated by 30 second cooling down intervals. DNA and RNA in the soluble sonicate were further degraded by adding 2 μl Benzonase (Novagen) and incubating at 4°C for 30 minutes. Cell debris was removed by centrifugation at 20,000g, 4°C for 30 minutes.

Inverse phase transition was induced by adding NaCl to the cell lysate at room temperature to achieve a final concentration of 2.0 M, followed by centrifugation at 20,000g for 15 minutes at room temperature. The resulting pellet contained the respective ELP-AR-LBD fusion protein and non-specifically NaCl precipitated proteins.

The pellet was re-suspended in 40 ml ice-cold 50mM Hepes pH 7.5, 150 mM NaCl, 0.1% N-Octylglycoside, 10% glycerol, 1 mM DTT and 1 μM DHT and re-centrifuged at 20,000 g, 4°C for 15 minutes to remove the non-specifically NaCl precipitated proteins. The inverse transition

cycle was repeated two additional times to increase the purity of the respective ELP-AR-LBD fusion protein and reduce the final volume to 25 ml.

(G) Isolation and Purification of Fusion Protein Containing Glucocorticoid Receptor Ligand Binding Domain (GR-LBD)

A single colony of *E. coli* strain BL21 Star (DE3) containing the ELP-GR-LBD fusion protein was inoculated into 500 ml CircleGrow (Q-BIOgene, San Diego, CA) supplemented with 100 μg/ml ampicillin (Sigma) and grown at 37°C with shaking at 250 rpm for 24 hours. The culture was harvested and suspended in 50 mM Hepes pH 7.5, 150 mM NaCl, 1 mM DTT, 10% glycerol, 0.1% CHAPS and 1 Complete EDTA free Protease inhibitor pellet (Roche, Indianapolis, IN). Cells were lysed by ultrasonic disruption on ice for 3 minutes, which consisted of 10 seconds bursts at 35% power separated by 30 second cooling down intervals. DNA and RNA in the soluble lysate were further degraded by adding 2μl Benzonase (Novagen) and incubating at 4°C for 30 minutes. Cell debris was removed by centrifugation at 20,000g, 4°C for 30 minutes.

Inverse phase transition was induced by adding NaCl to the cell lysate at room temperature to achieve a final concentration of 2.0 M, followed by centrifugation at 20,000g for 15 minutes at room temperature. The resulting pellet contained the ELP-GR-LBD fusion protein and non-specifically NaCl precipitated proteins.

The pellet was re-suspended in 40 ml ice-cold in 50 mM Hepes pH 7.5, 150 mM NaCl, 1 mM DTT, 10% glycerol, 0.1% CHAPS and re-centrifuged at 20,000 g, 4°C for 15 minutes to remove the non-specifically NaCl precipitated proteins. The inverse transition cycle was repeated two

additional times to increase the purity of the ELP-GR-LBD fusion protein and reduce the final volume to 10 ml.

(H) Isolation and Purification of Fusion Proteins Containing Estrogen Receptor Ligand Binding Domain (ERα-LBD)

A single colony of *E. coli* strain BL21 Star (DE3) containing the respective ELP-ERα-LBD fusion protein was inoculated into 500 ml CircleGrow (Q-BIOgene, San Diego, CA) supplemented with 100 μg/ml ampicillin (Sigma), 10% sucrose (Sigma) and grown at 27°C with shaking at 250 rpm for 48 hours. The culture was harvested and suspended in 40 ml 50mM Tris-HCL pH 8.0, 150 mM KCL, 1 mM EDTA, 1 mM DTT and 1 Complete EDTA free Protease inhibitor pellet (Roche, Indianapolis, IN). Cells were lysed by ultrasonic disruption on ice for 3 minutes, which consisted of 10 seconds bursts at 35% power separated by 30 second cooling down intervals. DNA and RNA in the soluble lysate were further degraded by adding 2 μl Benzonase (Novagen) and incubating at 4°C for 30 minutes. Cell debris was removed by centrifugation at 20,000g, 4°C for 30 minutes.

Inverse phase transition was induced by adding NaCl to the cell lysate at room temperature to achieve a final concentration of 1.5 M, followed by centrifugation at 20,000g for 15 minutes at room temperature. The resulting pellet contained the respective ELP-ERα-LBD fusion protein and non-specifically NaCl precipitated proteins.

The pellet was re-suspended in 40 ml ice-cold 50mM Tris-HCL pH 8.0, 150 mM KCL, 1 mM EDTA, 1 mM DTT and re-centrifuged at 20,000 g, 4°C for 15 minutes to remove the non-specifically NaCl precipitated proteins. The inverse transition cycle was repeated two additional

times to increase the purity of the respective ELP-ERα-LBD fusion protein and reduce the final volume to 10 ml.

(I) Isolation and Purification of Fusion Proteins Containing G Protein Alpha Q (Gαq)

A single colony of *E. coli* strain BL21 Star (DE3) containing the respective ELP-$G_{\alpha q}$ fusion protein was inoculated into 500 ml CircleGrow (Q-BIOgene, San Diego, CA) supplemented with 100 μg/ml ampicillin (Sigma) and 1 μM GDP and grown at 37°C with shaking at 250 rpm for 24 hours. The culture was harvested and suspended in 40 ml 50mM Hepes pH 7.5, 150 mM NaCl, 1.0% CHAPS, 10% glycerol, 1 mM DTT, 10 μM GDP and 1 Complete EDTA free Protease inhibitor pellet (Roche, Indianapolis, IN). Cells were lysed by ultrasonic disruption on ice for 3 minutes, which consisted of 10 seconds bursts at 35% power separated by 30 second cooling down intervals. DNA and RNA in the soluble lysate were further degraded by adding 2 μl Benzonase (Novagen) and incubating at 4°C for 30 minutes. Cell debris was removed by centrifugation at 20,000g, 4°C for 30 minutes.

Inverse phase transition was induced by adding NaCl to the cell lysate at room temperature to achieve a final concentration of 2.0 M, followed by centrifugation at 20,000g for 15 minutes at room temperature. The resulting pellet contained the respective ELP-$G_{\alpha q}$ fusion protein and non-specifically NaCl precipitated proteins.

The pellet was re-suspended in 30 ml ice-cold 50mM Hepes pH 7.5, 150 mM NaCl, 1.0% CHAPS, 10% glycerol, 1 mM DTT, 10 μM GDP and re-centrifuged at 20,000 g, 4°C for 15 minutes to remove the non-specifically NaCl precipitated proteins. The inverse transition cycle

was repeated two additional times to increase the purity of the respective ELP-$G_{\alpha q}$ fusion protein and reduce the final volume to 5 ml.

(J) Isolation and Purification of Fusion Proteins Containing 1-Deoxy-D-Xylulose 5-Phosphate Reductoisomerase (DXR)

A single colony of *E. coli* strain BL21 Star (DE3) containing the respective ELP-DXR fusion protein was inoculated into 500 ml CircleGrow (Q-BIOgene, San Diego, CA) supplemented with 100 μg/ml ampicillin (Sigma), 1mM $MnCl_2$ (VWR) and grown at 37°C with shaking at 250 rpm for 24 hours. The culture was harvested and suspended in 40 ml 0.1M Tris pH 7.6, 1 mM DTT and 1 Complete EDTA free Protease inhibitor pellet (Roche, Indianapolis, IN). Cells were lysed by ultrasonic disruption on ice for 3 minutes, which consisted of 10 seconds bursts at 35% power separated by 30 second cooling down intervals. DNA and RNA in the soluble lysate were further degraded by adding 2 μl Benzonase (Novagen) and incubating at 4°C for 30 minutes. Cell debris was removed by centrifugation at 20,000g at 4°C for 30 minutes.

Inverse phase transition was induced by adding NaCl to the cell lysate at room temperature to achieve a final concentration of 2.0 M, followed by centrifugation at 20,000g for 15 minutes at room temperature. The resulting pellet contained the respective ELP-DXR fusion protein and non-specifically NaCl precipitated proteins.

The pellet was re-suspended in 20 ml ice-cold 0.1 M Tris pH7.6, 1mM DTT and centrifuged at 20,000 g, 4°C for 15 minutes to remove the non-specifically NaCl precipitated proteins. The inverse transition cycle was repeated two additional times to increase the purity of the respective ELP-DXR fusion protein and reduce the final volume to 5 ml.

(K) Isolation and Purification of Fusion Protein Containing G Protein Alpha S (Gαs)

A single colony of *E. coli* strain BL21 Star (DE3) containing the ELP-$G_{\alpha s}$ fusion protein was inoculated into 500 ml CircleGrow (Q-BIOgene, San Diego, CA) supplemented with 100 μg/ml ampicillin (Sigma) and grown at 37°C with shaking at 250 rpm for 24 hours. The culture was harvested and suspended in 40 ml PBS, 10% glycerol, 1 mM DTT and 1 Complete EDTA free Protease inhibitor pellet (Roche, Indianapolis, IN). Cells were lysed by ultrasonic disruption on ice for 3 minutes, which consisted of 10 seconds bursts at 35% power separated by 30 second cooling down intervals. DNA and RNA in the soluble lysate were further degraded by adding 2 μl Benzonase (Novagen) and incubating at 4°C for 30 minutes. Cell debris was removed by centrifugation at 20,000g, 4°C for 30 minutes.

Inverse phase transition was induced by adding NaCl to the cell lysate at room temperature to a final concentration of 1.5 M, followed by centrifugation at 20,000g for 15 minutes at room temperature. The resulting pellet contained the ELP-$G_{\alpha s}$ fusion protein and non-specifically NaCl precipitated proteins.

The pellet was re-suspended in 10 ml ice-cold PBS, 10% glycerol, 1 mM DTT and centrifuged at 20,000 g, 4°C for 15 minutes to remove the non-specifically NaCl precipitated proteins. The inverse transition cycle was repeated two additional times to increase the purity of the ELP-$G_{\alpha s}$ fusion protein and reduce the final volume to 1 ml.

(11) THAT variations of the above-listed target proteins in their primary structures, secondary structures, tertiary structures, molecular weights, electrical charge distributions, viscosity, and biological functions,

did not prohibit the respective ELP-containing fusion proteins from retaining the inverse phase transition behavior of the ELPs.

Dr. Susan Dagher

2-27-04
Date

Dr. Susan Dagher appeared before me on this 27 day of February, 2004. She declared to me that she is the person described in this Affidavit, and she executed this Affidavit before me, and declared that her execution was completely voluntary.

State of North Carolina

County of Orange

"OFFICIAL SEAL"
Notary Public, North Carolina
County of Orange
Yongzhi Yang
My Commission Expires 10/10/2005

(SEAL)

Notary Public

My commission expires: 10/10/2005

Appendix A: Fusion Proteins

FP #	ELP Construct			Protease Cleavage Site in Spacer	Target Protein Construct		
	ELP Identification	Amino Acid #	Molecular Weight		Target Peptide/Protein	Amino Acid #	Molecular Weight
1	ELP4-60	301	24.6 KDa	Enterokinase protease (DDDDK)	Insulin A Peptide	21	2.4 KDa
2	ELP1-90	451	35.2 KDa	Enterokinase protease (DDDDK)	Insulin A Peptide	21	2.4 KDa
3	ELP4-120	601	49.2 KDa	Enterokinase protease (DDDDK)	Insulin A Peptide	21	2.4 KDa
4	ELP1-180	901	70.4 KDa	Enterokinase protease (DDDDK)	Insulin A Peptide	21	2.4 KDa
5	ELP4-60	301	24.6 KDa	Enterokinase protease (DDDDK)	T20 Peptide	36	4.4 KDa
6	ELP1-90	451	35.2 KDa	Enterokinase protease (DDDDK)	T20 Peptide	36	4.4 KDa
7	ELP4-120	601	49.2 KDa	Enterokinase protease (DDDDK)	T20 Peptide	36	4.4 KDa
8	ELP4-60	301	24.6 KDa	Thrombin protease (R/G)	T20 Peptide	36	4.4 KDa
9	ELP1-90	451	35.2 KDa	Thrombin protease (R/G)	T20 Peptide	36	4.4 KDa
10	ELP4-120	601	49.2 KDa	Thrombin protease (R/G)	T20 Peptide	36	4.4 KDa
11	ELP4-60	301	24.6 KDa	TEV protease (Q/S)	T20 Peptide	36	4.4 KDa
12	ELP1-90	451	35.2 KDa	TEV protease (Q/S)	T20 Peptide	36	4.4 KDa
13	ELP4-120	601	49.2 KDa	TEV protease (Q/S)	T20 Peptide	36	4.4 KDa
14	ELP4-60	301	24.6 KDa	TEV protease (Q/Y)	T20 Peptide	36	4.4 KDa
15	ELP1-90	451	35.2 KDa	TEV protease (Q/Y)	T20 Peptide	36	4.4 KDa
16	ELP4-120	601	49.2 KDa	TEV protease (Q/Y)	T20 Peptide	36	4.4 KDa
17	ELP1-90	451	35.2 KDa	Thrombin protease (R/G)	Interferon Alpha 2B	188	21.5 KDa
18	ELP4-60	301	24.6 KDa	Thrombin protease (R/G)	Tobacco Etch Virus Protease	242	27.5 KDa
19	ELP1-90	451	35.2 KDa	Thrombin protease (R/G)	Tobacco Etch Virus Protease	242	27.5 KDa
20	ELP4-120	601	49.2 KDa	Thrombin protease (R/G)	Tobacco Etch Virus Protease	242	27.5 KDa
21	ELP1-180	901	70.4 KDa	Thrombin protease (R/G)	Tobacco Etch Virus Protease	242	27.5 KDa
22	ELP1-90	451	35.2 KDa	Thrombin protease (R/G)	Small Heterodimer Partner Orphan Receptor	257	28.0 KDa
23	ELP1-90	451	35.2 KDa	Thrombin protease (R/G)	Androgen Receptor Ligand Binding Domain	258	30.1 KDa

24	ELP1-180	901	70.4 KDa	Thrombin protease (R/G)	Androgen Receptor Ligand Binding Domain	258	30.1 KDa
25	ELP1-90	451	35.2 KDa	Thrombin protease (R/G)	Glucocorticoid Receptor Ligand Binding Domain	279	32.1 KDa
26	ELP1-60	301	23.5 KDa	Thrombin protease (R/G)	Estrogen Receptor Ligand Binding Domain	296	33.2 KDa
27	ELP1-90	451	35.2 KDa	Thrombin protease (R/G)	Estrogen Receptor Ligand Binding Domain	296	33.2 KDa
28	ELP1-180	901	70.4 KDa	Thrombin protease (R/G)	Estrogen Receptor Ligand Binding Domain	296	33.2 KDa
29	ELP1-90	451	35.2 KDa	TEV protease (Q/G)	Estrogen Receptor Ligand Binding Domain	296	33.2 KDa
30	ELP1-90	451	35.2 KDa	Thrombin protease (R/G)	G Protein Alpha Q	359	42.1 KDa
31	ELP1-180	901	70.4 KDa	Thrombin protease (R/G)	G Protein Alpha Q	359	42.1 KDa
32	ELP1-60	301	23.5 KDa	Thrombin protease (R/G)	1-Deoxy-D-Xylulose 5-Phosphate Reductoisomerase Peptide	400	43.5 KDa
33	ELP1-90	451	35.2 KDa	Thrombin protease (R/G)	1-Deoxy-D-Xylulose 5-Phosphate Reductoisomerase Peptide	400	43.5 KDa
34	ELP1-180	901	70.4 KDa	Thrombin protease (R/G)	1-Deoxy-D-Xylulose 5-Phosphate Reductoisomerase Peptide	400	43.5 KDa
35	ELP1-90	451	35.2 KDa	TEV protease (Q/G)	1-Deoxy-D-Xylulose 5-Phosphate Reductoisomerase Peptide	400	43.5 KDa
36	ELP1-90	451	35.2 KDa	Thrombin protease (R/G)	G Protein Alpha S	380	44.2 KDa

Appendix B: Protein Sequences

(A). Target Protein Sequences

(1) Insulin A Peptide:

GIVEQCCTSICSLYQLENYCN

(2) T20 Peptide:

YTSLIHSLIEESQNQQEKNEQELLELDKWASLWNWF

(3) Interferon Alpha 2B:

MALTFALLVALLVLSCKSSCSVGCDLPQTHSLGSRRTLMLLAQMRRISLFSCLKDRHDFGFPQEEFGNQF
QKAETIPVLHEMIQQIFNLFSTKDSSAAWDETLLDKFYTELYQQLNDLEACVIQGVGVTETPLMKEDSIL
AVRKYFQRITLYLKEKKYSPCAWEVVRAEIMRSFSLSTNLQESLRSKE

(4) Tobacco Etch Virus Protease:

GESLFKGPRDYNPISSTICHLTNESDGHTTSLYGIGFGPFIITNKHLFRRNNGTLLVQSLHGVFKVKNTT
TLQQHLIDGRDMIIIRMPKDFPPFPQKLKFREPQREERICLVTTNFQTKSMSSMVSDTSCTFPSSDGIFW
KHWIQTKDGQCGSPLVSTRDGFIVGIHSASNFTNTNNYFTSVPKNFMELLTNQEAQQWVSGWRLNADSVL
WGGHKVFMSKPEEPFQPVKEATQLMNELVYSQ

(5) Small Heterodimer Partner Orphan Receptor:

MSTSQPGACPCQGAASRPAILYALLSSSLKAVPRPRSRCLCRQHRPVQLCAPHRTCREALDVLAKTVAFL
RNLPSFWQLPPQDQRRLLQGCWGPLFLLGLAQDAVTFEVAEAPVPSILKKILLEEPSSSGGSGQLPDRPQ
PSLAAVQWLQCCLESFWSLELSPKEYACLKGTILFNPDVPGLQAASHIGHLQQEAHWVLCEVLEPWCPAA
QGRLTRVLLTASTLKSIPTSLLGDLFFRPIIGDVDIAGLLGDMLLLR

(6) Androgen Receptor Ligand Binding Domain:

MHIEGYECQPIFLNVLEAIEPGVVCAGHDNNQPDSFAALLSSLNELGERQLVHVVKWAKALPGFRNLHVD
DQMAVIQYSWMGLMVFAMGWRSFTNVNSRMLYFAPDLVFNEYRMHKSRMYSQCVRMRHLSQEFGWLQITP
QEFLCMKALLLFSIIPVDGLKNQKFFDELRMNYIKELDRIIACKRKNPTSCSRRFYQLTKLLDSVQPIAR
ELHQFTFDLLIKSHMVSVDFPEMMAEIISVQVPKILSGKVKPIYFHTQ

(7) Glucocorticoid Receptor Ligand Binding Domain:

MIQQATTGVSQETSENPGDKTIVPATLPQLTPTLVSLLEVIEPEVLYAGYDSSVPDSTWRIMTTLNMLGG
RQVIAAVKWAKAIPGFRNLHLDDQMTLLQYSWMSLMAFALGWRSYRQSSANLLCFAPDLIINEQRMTLPD
MYDQCKHMLYVSSELHRLQVSYEEYLCMKTLLLLSSVPKDGLKSQELFDEIRMTYIKELGKAIVKREGNS
SQNWQRFYQLTKLLDSMHEVVENLLNYCFQTFLDKTMSIEFPEMLAEIITNQIPKYSNGNIKKLLFHQK

(8) Estrogen Receptor Ligand Binding Domain:

MSKKNSLALSLTADQMVSALLDAEPPILYSEYDPTRPFSEASMMGLLTNLADRELVHMINWAKRVPGFVD
LTLHDQVHLLECAWLEILMIGLVWRSMEHPGKLLFAPNLLLDRNQGKCVEGMVEIFDMLLATSSRFRMMN
LQGEEFVCLKSIILLNSGVYTFLSSTLKSLEEKDHIHRVLDKITDTLIHLMAKAGLTLQQQHQRLAQLLL
ILSHIRHMSNKGMEHLYSMKCKNVVPLYDLLLEMLDAHRLHAPTSRGGASVEETDQSHLATAGSTSSHSL
QKYYITGEAEGFPATV

(9) G Protein Alpha Q:

MTLESIMACCLSEEAKEARRINDEIERQLRRDKRDARRELKLLLLGTGESGKSTFIKQMRIIHGSGYSDE
DKRGFTKLVYQNIFTAMQAMIRAMDTLKIPYKYEHNKAHAQLVREVDVEKVSAFENPYVDAIKSLWNDPG
IQECYDRRREYQLSDSTKYYLNDLDRVADPAYLPTQQDVLRVRVPTTGIIEYPFDLQSVIFRMVDVGGQR
SERRKWIHCFENVTSIMFLVALSEYDQVLVESDNENRMEESKALFRTIITYPWFQNSSVILFLNKKDLLE
EKIMYSHLVDYFPEYDGPQRDAQAAREFILKMFVDLNPDSDKINYSHFTCATDTENIRFVFAAVKDTILQ
LNLKEYNLV

(10) 1-Deoxy-D-Xylulose 5-Phosphate Reductoisomerase Peptide:

MKQLTILGSTGSIGCSTLDVVRHNPEHFRVVALVAGKNVTRMVEQCLEFSPRYAVMDDEASAKLLKTMLQ
QQGSRTEVLSGQQAACDMAALEDVDQVMAAIVGAAGLLPTLAAIRAGKTILLANKESLVTCGRLFMDAVK
QSKAQLLPVDSEHNAIFQSLPQPIQHNLGYADLEQNGVVSILLTGSGGPFRETPLRDLATMTPDQACRHP
NWSMGRKISVDSATMMNKGLEYIEARWLFNASASQMEVLIHPQSVIHSMVRYQDGSVLAQLGEPDMRTPI
AHTMAWPNRVNSGVKPLDFCKLSALTFAAPDYDRYPCLKLAMEAFEQGQAATTALNAANEITVAAFLAQQ
IRFTDIAALNLSVLEKMDMREPQCVDDVLSVDASAREVARKEVMRLASPV

(11) G Protein Alpha S:

MGCLGNSKTEDQRNEEKAQREANKKIEKQLQKDKQVYRATHRLLLLGAGESGKSTIVKQMRILHVNGFNG
DSEKATKVQDIKNNLKEAIETIVAAMSNLVPPVELANPENQFRVDYILSVMNVPDFDFPPEFYEHAKALW
EDEGVRACYERSNEYQLIDCAQYFLDKIDVIKQADYVPSDQDLLRCRVLTSGIFETKFQVDKVNFHMFDV
GGQRDERRKWIQCFNDVTAIIFVVASSSYNMVIREDNQTNRLQEALNLFKSIWNNRWLRTISVILFLNKQ
DLLAEKVLAGKSKIEDYFPEFARYTTPEDATPEPGEDPRVTRAKYFIRDEFLRISTASGDGRHYCYPHFT
CAVDTENIRRVFNDCRDIIQRMHLRQYELL

(B). Phase Transition Protein Sequences

(1) ELP1-60 (PflM1-Bgl1):

GVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVP
GVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVP
GAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVP
GGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVP
GVGVPGGGVPGAGVPGGGVPG

(2) ELP4-60 (PflM1-Bgl1):

GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP
GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP
GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP

GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP
GVGVPGVGVPGVGVPGVGVPG

(3) <u>ELP1-90 (PflM1-Bgl1)</u>:

GVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVP
GVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVP
GAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVP
GGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVP
GVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVP
GVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVP
GVGVPGVGVPGVGVPGGGVPGAGVPGGGVPG

(4) <u>ELP4-120 (PflM1-Bgl1)</u>:

GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP
GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP
GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP
GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP
GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP
GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP
GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP
GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVP
GVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG

(5) <u>ELP1-180 (PflM1-Bgl1)</u>:

GVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVP
GVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVP
GAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVP
GGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVP
GVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVP
GVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVP
GVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVP
GAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVP
GGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVP
GVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVP
GVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVP
GVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVP
GAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPG

<u>(C). Spacer Sequences</u>

(1) <u>Containing thrombin protease (throm) cleavage site R/G</u>:

(a) WPSSGGGGGSIGPLVPRGSH

(b) WPSSGLVPRGSPGISGGGGGH

(c) WPSSGGGSIGPLVPRGSHSMGLNDIFEAQKIEWHEHMPMALE

(d) WPSSGGGGGSIGPLVPRGSHMPMALE

(e) WPGASGGGGPLVPRGS

(f) WPSSGLVPRGSPGISGGGGGHMP

(g) WPSSGLVPRGSPGISGGGGGHMPM

(2) Containing enterokinase protease (EK) cleavage site K/G:

(a) WPGASSGTDDDDK

(3) Containing tobacco etch virus (TEV) protease cleavage site Q/S:

(a) WPGASGPTTENLYFQS

(4) Containing tobacco etch virus protease (TEV) cleavage site Q/Y:

(a) WPGASGPTTENLYFQK----

Protein

(5) Containing tobacco etch virus (TEV) protease cleavage site Q/G:

(a) WPSSGDYDIPTTENLYFQGAH

(D) Fusion Protein Sequences[1]

(1) FP#1: (pET32a-SD15-ELP4-60-EK-Insulin A peptide):

MGGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPG**WPGASSGTDDDDK/GIVEQCCTSICSLYQLENYCN**

(2) FP#2: (pET32a-SD15-ELP1-90-EK-Insulin A peptide):

MGGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPG
GGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPG
VGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPG
VGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPG

[1] with ELP protein underlined, target protein bold and underlined, spacer marked by gray, and cleavage site indicated by a forward slash.

GGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPGASSGTDDDDK/**GIVEQCCTSICSLYQLENYCN**

(3) FP#3: (pET32a-SD15-ELP4-120-EK-Insulin A peptide):

MGGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGWPGASSGTDDDDK/**GIVEQCCTSIC**
SLYQLENYCN

(4) FP#4: (pET32a-SD15-ELP1-180-EK-Insulin A peptide):

MGGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPG
GGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPG
VGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPG
VGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPG
GGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPG
GGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPG
VGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPG
VGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPG
GGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPG
GGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPGAS
SGTDDDDK/**GIVEQCCTSICSLYQLENYCN**

(5) FP#5: (pET15b-ELP4-60-EK-T20 peptide):

MGGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGWPGASSGTDDDDK/**YTSLIHSLIEESQNQQEKNEQELLELDKWAS**
LWNWF

(6) FP#6: (pET17b-ELP1-90-EK-T20 peptide):

MGGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPG
GGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPG
VGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPG

VGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPG
GGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPGASSGTDDDDK/**YTSLIHSLIEESQNQQEKNEQ**
ELLELDKWASLWNWF

(7) FP#7: (pET15b-ELP4-120-EK-T20 peptide):

MGGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGWPGASSGTDDDDK/**YTSLIHSLIEE**
SQNQQEKNEQELLELDKWASLWNWF

(8) FP#8: (pET17b-ELP4-60-Throm-T20 peptide):

MGGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGWPGASGGGGPLVPR/GS**YTSLIHSLIEESQNQQEKNEQELLELDK**
WASLWNWF

(9) FP#9: (pET17b-ELP1-90-Throm-T20 peptide):

MGGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPG
GGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPG
VGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPG
VGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPG
GGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPGASGGGGPLVPR/GS**YTSLIHSLIEESQNQQEK**
NEQELLELDKWASLWNWF

(10) FP#10: (pET17b-ELP4-120-Throm-T20 peptide):

MGGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG

VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGWPGASGGGGPLVPR/GS**YTSLIHSL**
IEESQNQQEKNEQELLELDKWASLWNWF

(11) FP#11:(pET17b-ELP4-60-TEV(Q/S)-T20 peptide):

MGGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGWPGASGPTTENLYFQ/S**YTSLIHSLIEESQNQQEKNEQELLELDK**
WASLWNWF

(12) FP#12: (pET17b-ELP1-90-TEV(Q/S)-T20 peptide):

MGGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPG
GGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPG
VGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPG
VGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPG
GGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPGASGPTTENLYFQ/S**YTSLIHSLIEESQNQQEK**
NEQELLELDKWASLWNWF

(13) FP#13: (pET17b-ELP4-120-TEV(Q/S)-T20 peptide):

MGGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGWPGASGPTTENLYFQ/S**YTSLIHSL**
IEESQNQQEKNEQELLELDKWASLWNWF

(14) FP#14: (pET17b-ELP4-60-TEV(Q/Y)-T20 peptide):

MGGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGWPGASGPTTENLYFQ/**YTSLIHSLIEESQNQQEKNEQELLELDKW**
ASLWNWF

(15) FP#15: (pET17b-ELP1-90-TEV(Q/Y)-T20 peptide):

MGGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPG

GGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPG
VGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPG
VGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPG
GGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPG
AGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPGASGPTTENLYFQ/**YTSLIHSLIEESQNQQEKN**
EQELLELDKWASLWNWF

(16) FP#16: (pET17b-ELP4-120-TEV(Q/Y)-T20 peptide):

MGGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG
VGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG**WPGASGPTTENLYFQ/YTSLIHSLI**
EESQNQQEKNEQELLELDKWASLWNWF

(17) FP#17: (pET32a-SD11-ELP1-90-throm-Interferon Alpha 2B):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPG**WPSSGLVPR/GSPGISGGGGGHMPMALTFALL**
VALLVLSCKSSCSVGCDLPQTHSLGSRRTLMLLAQMRRISLFSCLKDRHDFGFPQEEFGNQFQKAETIPV
LHEMIQQIFNLFSTKDSSAAWDETLLDKFYTELYQQLNDLEACVIQGVGVTETPLMKEDSILAVRKYFQR
ITLYLKEKKYSPCAWEVVRAEIMRSFSLSTNLQESLRSKE

(18) FP#18: (pET15b-SD5-ELP4-60-throm-Tobacco etch virus protease):

MRALMGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPG**WPSSGLVPR/GSPGISGGGGGHMPMGESLFKGPRDYNPISST**
ICHLTNESDGHTTSLYGIGFGPFIITNKHLFRRNNGTLLVQSLHGVFKVKNTTTLQQHLIDGRDMIIIRM
PKDFPPFPQKLKFREPQREERICLVTTNFQTKSMSSMVSDTSCTFPSSDGIFWKHWIQTKDGQCGSPLVS
TRDGFIVGIHSASNFTNTNNYFTSVPKNFMELLTNQEAQQWVSGWRLNADSVLWGGHKVFMSKPEEPFQP
VKEATQLMNELVYSQ

(19) FP#19: (pET15b-SD5-ELP1-90-throm-Tobacco etch virus protease):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG

VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPSSGLVPR/GSPGISGGGGGHMPM**GESLFKG**
PRDYNPISSTICHLTNESDGHTTSLYGIGFGPFIITNKHLFRRNNGTLLVQSLHGVFKVKNTTTLQQHLI
DGRDMIIIRMPKDFPPFPQKLKFREPQREERICLVTTNFQTKSMSSMVSDTSCTFPSSDGIFWKHWIQTK
DGQCGSPLVSTRDGFIVGIHSASNFTNTNNYFTSVPKNFMELLTNQEAQQWVSGWRLNADSVLWGGHKVF
MSKPEEPFQPVKEATQLMNELVYSQ

(20) FP#20: (pET15b-SD5-ELP4-120-throm-Tobacco etch virus protease):

MRALMGPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVG
VPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGVGVPGWPSSGLVPR/GSPGISGGGGGH
MPM**GESLFKGPRDYNPISSTICHLTNESDGHTTSLYGIGFGPFIITNKHLFRRNNGTLLVQSLHGVFKVK**
NTTTLQQHLIDGRDMIIIRMPKDFPPFPQKLKFREPQREERICLVTTNFQTKSMSSMVSDTSCTFPSSDG
IFWKHWIQTKDGQCGSPLVSTRDGFIVGIHSASNFTNTNNYFTSVPKNFMELLTNQEAQQWVSGWRLNAD
SVLWGGHKVFMSKPEEPFQPVKEATQLMNELVYSQ

(21) FP#21: (pET15b-SD5-ELP1-180-throm-Tobacco etch virus protease):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWP
SSGLVPR/GSPGISGGGGGHMPM**GESLFKGPRDYNPISSTICHLTNESDGHTTSLYGIGFGPFIITNKHL**
FRRNNGTLLVQSLHGVFKVKNTTTLQQHLIDGRDMIIIRMPKDFPPFPQKLKFREPQREERICLVTTNFQ
TKSMSSMVSDTSCTFPSSDGIFWKHWIQTKDGQCGSPLVSTRDGFIVGIHSASNFTNTNNYFTSVPKNFM
ELLTNQEAQQWVSGWRLNADSVLWGGHKVFMSKPEEPFQPVKEATQLMNELVYSQ

(22) FP#22: (pET15b-SD3-ELP1-90-throm-Small Heterodimer partner orphan receptor):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPG**WPSSGGGGGSIGPLVPR/GSHMSTSQPGACPC**
QGAASRPAILYALLSSSLKAVPRPRSRCLCRQHRPVQLCAPHRTCREALDVLAKTVAFLRNLPSFWQLPP
QDQRRLLQGCWGPLFLLGLAQDAVTFEVAEAPVPSILKKILLEEPSSSGGSGQLPDRPQPSLAAVQWLQC
CLESFWSLELSPKEYACLKGTILFNPDVPGLQAASHIGHLQQEAHWVLCEVLEPWCPAAQGRLTRVLLTA
STLKSIPTSLLGDLFFRPIIGDVDIAGLLGDMLLLR

(23) FP#23: (pET15b-SD3-ELP1-90-throm-Androgen receptor ligand binding domain):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPG**WPSSGGGGGSIGPLVPR/GSHMHIEGYECQPI**
FLNVLEAIEPGVVCAGHDNNQPDSFAALLSSLNELGERQLVHVVKWAKALPGFRNLHVDDQMAVIQYSWM
GLMVFAMGWRSFTNVNSRMLYFAPDLVFNEYRMHKSRMYSQCVRMRHLSQEFGWLQITPQEFLCMKALLL
FSIIPVDGLKNQKFFDELRMNYIKELDRIIACKRKNPTSCSRRFYQLTKLLDSVQPIARELHQFTFDLLI
KSHMVSVDFPEMMAEIISVQVPKILSGKVKPIYFHTQ

(24) FP#24: (pET15b-SD3-ELP1-180-throm-Androgen receptor ligand binding domain):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWP
SSGGGGGSIGPLVPR/GSHMHIEGYECQPIFLNVLEAIEPGVVCAGHDNNQPDSFAALLSSLNELGERQL
VHVVKWAKALPGFRNLHVDDQMAVIQYSWMGLMVFAMGWRSFTNVNSRMLYFAPDLVFNEYRMHKSRMYS
QCVRMRHLSQEFGWLQITPQEFLCMKALLLFSIIPVDGLKNQKFFDELRMNYIKELDRIIACKRKNPTSC
SRRFYQLTKLLDSVQPIARELHQFTFDLLIKSHMVSVDFPEMMAEIISVQVPKILSGKVKPIYFHTQ

(25) FP#25: (pET15b-SD3-ELP1-90-throm-Glucocorticoid receptor ligand binding domain):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPG**WPSSGGGGGSIGPLVPR/GSHMIQQATTGVSQ**
ETSENPGDKTIVPATLPQLTPTLVSLLEVIEPEVLYAGYDSSVPDSTWRIMTTLNMLGGRQVIAAVKWAK
AIPGFRNLHLDDQMTLLQYSWMSLMAFALGWRSYRQSSANLLCFAPDLIINEQRMTLPDMYDQCKHMLYV
SSELHRLQVSYEEYLCMKTLLLLSSVPKDGLKSQELFDEIRMTYIKELGKAIVKREGNSSQNWQRFYQLT
KLLDSMHEVVENLLNYCFQTFLDKTMSIEFPEMLAEIITNQIPKYSNGNIKKLLFHQK

(26) FP#26: (pET15b-SD3-ELP1-60-throm-Estrogen receptor ligand binding domain):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPG**WPSSGGGGGSIGPLVPR/GSHMSKKNSLALSLTADQMVSALL**
DAEPPILYSEYDPTRPFSEASMMGLLTNLADRELVHMINWAKRVPGFVDLTLHDQVHLLECAWLEILMIG
LVWRSMEHPGKLLFAPNLLLDRNQGKCVEGMVEIFDMLLATSSRFRMMNLQGEEFVCLKSIILLNSGVYT
FLSSTLKSLEEKDHIHRVLDKITDTLIHLMAKAGLTLQQQHQRLAQLLLILSHIRHMSNKGMEHLYSMKC
KNVVPLYDLLLEMLDAHRLHAPTSRGGASVEETDQSHLATAGSTSSHSLQKYYITGEAEGFPATV

(27) FP#27: (pET15b-SD5-ELP1-90-throm-Estrogen receptor ligand binding domain):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPG**WPSSGGGGGSIGPLVPR/GSHMSKKNSLALSL**
TADQMVSALLDAEPPILYSEYDPTRPFSEASMMGLLTNLADRELVHMINWAKRVPGFVDLTLHDQVHLLE
CAWLEILMIGLVWRSMEHPGKLLFAPNLLLDRNQGKCVEGMVEIFDMLLATSSRFRMMNLQGEEFVCLKS
IILLNSGVYTFLSSTLKSLEEKDHIHRVLDKITDTLIHLMAKAGLTLQQQHQRLAQLLLILSHIRHMSNK
GMEHLYSMKCKNVVPLYDLLLEMLDAHRLHAPTSRGGASVEETDQSHLATAGSTSSHSLQKYYITGEAEG
FPATV

(28) FP#28: (pET15b-SD5-ELP1-180-throm-Estrogen receptor ligand binding domain):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG

VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWP
SSGLVPR/GSPGISGGGGGHMSKKNSLALSLTADQMVSALLDAEPPILYSEYDPTRPFSEASMMGLLTNL
ADRELVHMINWAKRVPGFVDLTLHDQVHLLECAWLEILMIGLVWRSMEHPGKLLFAPNLLLDRNQGKCVE
GMVEIFDMLLATSSRFRMMNLQGEEFVCLKSIILLNSGVYTFLSSTLKSLEEKDHIHRVLDKITDTLIHL
MAKAGLTLQQQHQRLAQLLLILSHIRHMSNKGMEHLYSMKCKNVVPLYDLLLEMLDAHRLHAPTSRGGAS
VEETDQSHLATAGSTSSHSLQKYYITGEAEGFPATV

(29) FP#29: (pET15b-SD6-ELP1-90-TEV-Estrogen receptor ligand binding domain):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPSSGDYDIPTTENLYFQ/GAHMSKKNSLALS
LTADQMVSALLDAEPPILYSEYDPTRPFSEASMMGLLTNLADRELVHMINWAKRVPGFVDLTLHDQVHLL
ECAWLEILMIGLVWRSMEHPGKLLFAPNLLLDRNQGKCVEGMVEIFDMLLATSSRFRMMNLQGEEFVCLK
SIILLNSGVYTFLSSTLKSLEEKDHIHRVLDKITDTLIHLMAKAGLTLQQQHQRLAQLLLILSHIRHMSN
KGMEHLYSMKCKNVVPLYDLLLEMLDAHRLHAPTSRGGASVEETDQSHLATAGSTSSHSLQKYYITGEAE
GFPATV

(30) FP#30: (pET15b-SD1-ELP1-90-throm-G protein alpha Q):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPSSGGGSIGPLVPR/GSHSMGLNDIFEAQKI
EWHEHMPMALEMTLESIMACCLSEEAKEARRINDEIERQLRRDKRDARRELKLLLLGTGESGKSTFIKQM
RIIHGSGYSDEDKRGFTKLVYQNIFTAMQAMIRAMDTLKIPYKYEHNKAHAQLVREVDVEKVSAFENPYV
DAIKSLWNDPGIQECYDRRREYQLSDSTKYYLNDLDRVADPAYLPTQQDVLRVRVPTTGIIEYPFDLQSV
IFRMVDVGGQRSERRKWIHCFENVTSIMFLVALSEYDQVLVESDNENRMEESKALFRTIITYPWFQNSSV
ILFLNKKDLLEEKIMYSHLVDYFPEYDGPQRDAQAAREFILKMFVDLNPDSDKINYSHFTCATDTENIRF
VFAAVKDTILQLNLKEYNLV

(31) FP#31: (pET15b-SD1-ELP1-180-throm-G protein alpha Q):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG

VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWP
SSGGGSIGPLVPR/GSHSMGLNDIFEAQKIEWHEHMPMALE**MTLESIMACCLSEEAKEARRINDEIERQL**
RRDKRDARRELKLLLLGTGESGKSTFIKQMRIIHGSGYSDEDKRGFTKLVYQNIFTAMQAMIRAMDTLKI
PYKYEHNKAHAQLVREVDVEKVSAFENPYVDAIKSLWNDPGIQECYDRRREYQLSDSTKYYLNDLDRVAD
PAYLPTQQDVLRVRVPTTGIIEYPFDLQSVIFRMVDVGGQRSERRKWIHCFENVTSIMFLVALSEYDQVL
VESDNENRMEESKALFRTIITYPWFQNSSVILFLNKKDLLEEKIMYSHLVDYFPEYDGPQRDAQAAREFI
LKMFVDLNPDSDKINYSHFTCATDTENIRFVFAAVKDTILQLNLKEYNLV

(32) FP#32: (pET15b-SD3-ELP1-60-throm-1-Deoxy-D-Xylulose 5-Phosphate Reductoisomerase Peptide):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGWPSSGGGGGSIGPLVPR/GSH**MKQLTILGSTGSIGCSTLDVV**
RHNPEHFRVVALVAGKNVTRMVEQCLEFSPRYAVMDDEASAKLLKTMLQQQGSRTEVLSGQQAACDMAAL
EDVDQVMAAIVGAAGLLPTLAAIRAGKTILLANKESLVTCGRLFMDAVKQSKAQLLPVDSEHNAIFQSLP
QPIQHNLGYADLEQNGVVSILLTGSGGPFRETPLRDLATMTPDQACRHPNWSMGRKISVDSATMMNKGLE
YIEARWLFNASASQMEVLIHPQSVIHSMVRYQDGSVLAQLGEPDMRTPIAHTMAWPNRVNSGVKPLDFCK
LSALTFAAPDYDRYPCLKLAMEAFEQGQAATTALNAANEITVAAFLAQQIRFTDIAALNLSVLEKMDMRE
PQCVDDVLSVDASAREVARKEVMRLASPV

(33) FP#33: (pET15b-SD5-ELP1-90-throm-1-Deoxy-D-Xylulose 5-Phosphate Reductoisomerase Peptide):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPSSGLVPR/GSPGISGGGGGH**MKQLTILGST**
GSIGCSTLDVVRHNPEHFRVVALVAGKNVTRMVEQCLEFSPRYAVMDDEASAKLLKTMLQQQGSRTEVLS
GQQAACDMAALEDVDQVMAAIVGAAGLLPTLAAIRAGKTILLANKESLVTCGRLFMDAVKQSKAQLLPVD
SEHNAIFQSLPQPIQHNLGYADLEQNGVVSILLTGSGGPFRETPLRDLATMTPDQACRHPNWSMGRKISV
DSATMMNKGLEYIEARWLFNASASQMEVLIHPQSVIHSMVRYQDGSVLAQLGEPDMRTPIAHTMAWPNRV
NSGVKPLDFCKLSALTFAAPDYDRYPCLKLAMEAFEQGQAATTALNAANEITVAAFLAQQIRFTDIAALN
LSVLEKMDMREPQCVDDVLSVDASAREVARKEVMRLASPV

(34) FP#34: (pET15b-SD5-ELP1-180-throm-1-Deoxy-D-Xylulose 5-Phosphate Reductoisomerase Peptide):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVG
SSGLVPR/GSPGISGGGGGHMKQLTILGSTGSIGCSTLDVVRHNPEHFRVVALVAGKNVTRMVEQCLEFS
PRYAVMDDEASAKLLKTMLQQQGSRTEVLSGQQAACDMAALEDVDQVMAAIVGAAGLLPTLAAIRAGKTI
LLANKESLVTCGRLFMDAVKQSKAQLLPVDSEHNAIFQSLPQPIQHNLGYADLEQNGVVSILLTGSGGPF
RETPLRDLATMTPDQACRHPNWSMGRKISVDSATMMNKGLEYIEARWLFNASASQMEVLIHPQSVIHSMV
RYQDGSVLAQLGEPDMRTPIAHTMAWPNRVNSGVKPLDFCKLSALTFAAPDYDRYPCLKLAMEAFEQGQA
ATTALNAANEITVAAFLAQQIRFTDIAALNLSVLEKMDMREPQCVDDVLSVDASAREVARKEVMRLASPV

(35) FP#35: (pET15b-SD6-ELP1-90-TEV-1-Deoxy-D-Xylulose 5-Phosphate Reductoisomerase Peptide):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPSSGDYDIPTTENLYFQ/GAHMKQLTILGST
GSIGCSTLDVVRHNPEHFRVVALVAGKNVTRMVEQCLEFSPRYAVMDDEASAKLLKTMLQQQGSRTEVLS
GQQAACDMAALEDVDQVMAAIVGAAGLLPTLAAIRAGKTILLANKESLVTCGRLFMDAVKQSKAQLLPVD
SEHNAIFQSLPQPIQHNLGYADLEQNGVVSILLTGSGGPFRETPLRDLATMTPDQACRHPNWSMGRKISV
DSATMMNKGLEYIEARWLFNASASQMEVLIHPQSVIHSMVRYQDGSVLAQLGEPDMRTPIAHTMAWPNRV
NSGVKPLDFCKLSALTFAAPDYDRYPCLKLAMEAFEQGQAATTALNAANEITVAAFLAQQIRFTDIAALN
LSVLEKMDMREPQCVDDVLSVDASAREVARKEVMRLASPV

(36) FP#36: (pET15b-SD3-ELP1-90-throm-G protein alpha S):

MRALMGPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVG
VPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVG
VPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVG
VPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAG
VPGGGVPGVGVPGVGVPGGGVPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGVGVPGVGVPGGG
VPGAGVPGVGVPGVGVPGVGVPGGGVPGAGVPGGGVPGWPSSGGGGGSIGPLVPR/GSHMPMALEMGCLG
NSKTEDQRNEEKAQREANKKIEKQLQKDKQVYRATHRLLLLGAGESGKSTIVKQMRILHVNGFNGDSEKA
TKVQDIKNNLKEAIETIVAAMSNLVPPVELANPENQFRVDYILSVMNVPDFDFPPEFYEHAKALWEDEGV
RACYERSNEYQLIDCAQYFLDKIDVIKQADYVPSDQDLLRCRVLTSGIFETKFQVDKVNFHMFDVGGQRD
ERRKWIQCFNDVTAIIFVVASSSYNMVIREDNQTNRLQEALNLFKSIWNNRWLRTISVILFLNKQDLLAE

KVLAGKSKIEDYFPEFARYTTPEDATPEPGEDPRVTRAKYFIRDEFLRISTASGDGRHYCYPHFTCAVDT
ENIRRVFNDCRDIIQRMHLRQYELL